13 MICROSPECTROPHOTOMETRY (MSP)

A. QC Check

A QC check is performed prior to the analysis of any evidence samples and will include at a minimum the Holmium oxide and Didymium filters. If no casework has been performed in a given month, then the QC check will be performed. The neutral density filters will be included as a part of the QC check a minimum of once annually.

a. Turn on the microscope transmitted light lamp (rocker switch on back of microscope, right base near power plug). Slowly increase light intensity to maximum (green light stack totally lit). Note: field selector (BF, bright field, DF, dark field) should be set to DF (located at front of top illuminator).

b. Turn on the video monitor, the computer/monitor, and plotter. The sequence of individual instrument component start-up and shutdown can be random because the UV power source is not being used. Allow the optical system to warm up and stabilize for 30 minutes.

c. Place the clear Reference filter, from the filter set box, on the microscope stage and using the 20X objective, focus on the ink dot on the filter. Set Köhler illumination. Move to a clear area on the filter. Click AUTO GAIN on the top left of the desktop. The value obtained (maximum y counts) should range from 3700 to 4000, maximum saturation (values below 3700 are indicative of insufficient illumination). Click OK on the pop up menu. This step sets the integration time for the instrument.

d. Close the light path above the specimen by rotating the filter wheel from the white "O" position, to the next click (either side rotation). Video monitor image should be black. Click DARK SCAN (on left, under AUTO GAIN) to establish and adjust for instrument noise.

e. Open the light path above the specimen by rotating the filter wheel so the "O" is displayed and the light path is restored (light visible on the monitor). Take a "Reference Scan" which measures and compensates for the light transmitting/absorbing effect of the clear glass Reference filter, light source, optics, and later for the effect of the glass slide, coverslip, and mounting media in casework specimens. The effect of the room lighting, or stray light, is not detectable.

f. Place the Holmium oxide filter on top of the transmission field stop (below substage condenser) and take a SAMPLE SCAN (left desktop). Be sure to select %T, Choose OK. Choose the NIST (reference value) icon (third from top right end of the tool bar). Select Holmium oxide, VIS region, on pop up menu. Select "Mark all peaks" under "Peaks option". This displays the NIST filter specific certified values and the current SAMPLE SCAN peak values respectively. Choose FILE, print, OK, for hard copy with initials to be placed in the MSP QC notebook.

g. Repeat the above steps with the Didymium filter (A new screen is obtained by clicking the S.E.E icon at the far right of the tool bar and numbers from the previous scan can be removed by the option, Close all slots, under FILE). Print with the current measurement values and corresponding NIST certified Didymium filter peak values and initials. Place in MSP QC notebook.

h. Repeat the above steps for the three neutral density filters, as appropriate. Note: Choose ABSORBANCE instead of %T. When placing the neutral density filters on top of the transmission field stop be sure the filter label is on top and at your front left. These are the only filters that are orientation sensitive. Print with the current measurement values and corresponding NIST certified neutral density filter peak values and initials. Place in MSP QC notebook.

i. If the obtained values are not within ± 2 nm of the current NIST certified reference values, perform Köhler illumination adjustment on the microscope and repeat check of the instrument wavelength scale verification steps. **DO NOT PROCEED WITH CASEWORK SAMPLES** until the Holmium oxide and Didymium filter peak mark values are within ± 2 nm of the NIST lab-specific, certified reference values. Call S.E.E. Technical Support as needed.

j. The Holmium oxide, Didymium and neutral density filters were received with NIST certified values and both sets (Central and Western) were re-certified once. These filter sets will not be sent for re-certification on an annual basis as there is no expectation that their values will change if they are properly handled. The manufacturer allows a deviation of ± 4 nm from the certified values. Our procedure calls for ± 2 nm. The filter sets will only be sent for re-certification on an as needed basis.

B. Analysis of Casework Samples

a. Prior to using the MSP, evidence samples have already been visually and/or microscopically compared and found to have no significant, or unexplainable, differences in the side-by-side comparison of color. The QC check using the Holmium oxide and Didymium filters has also been completed prior to, and on the same day, as casework MSP spectra are obtained.

b. For fibers, the questioned and known samples must be mounted in the same media, e.g., water, xylene substitute, glycerol, or Permount. Ink and paint samples do not require mounting in a media.

c. Place the microscope slide containing the colored material on the stage and focus on a sample with the 20X or 50X objective depending upon the size of the colored material. The video monitor sampling aperture (black square) should be completely contained in the area of the sample to be measured. All light entering the square sampling aperture (seen on the video screen) must have been transmitted through the sample.

d. Move the sample away from the aperture area, until a reference (clear) area is found on the microscope slide. Run REFERENCE SCAN.

   i. Whenever the sample is moved out of the monitor field of view to another target location on the same or on a different slide, the REFERENCE SCAN function should be repeated.

e. Realign and focus the sample with reference to the video sampling aperture (black square image).

f. Collect a SAMPLE SCAN.

g. Each different slide used must have a REFERENCE SCAN taken prior to any SAMPLE SCANS.

h. Repeat AUTO GAIN and DARK SCAN periodically during lengthy sample measurement sessions.

i. Print and initial data, as needed for case file.

C. Shutdown Procedure

a. In order to preserve the life of the halogen bulb, **BEFORE** shutting off the lamp switch, slowly push the light intensity knob to the bottom.

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